NUTRITIONAL AND *IN VITRO* PROTEIN DIGESTIBILITY COMPARISON BETWEEN BEEF, HYBRID, AND PLANT-BASED ANALOGUE BURGERS

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I. INTRODUCTION

Meat alternatives, such as hybrid meat products and plant-based analogues, have been introduced to the market, often leaving consumers uncertain about their nutritional quality, which is directly affected by the ingredients in their formulations. Plant proteins are known to lack some essential amino acids and contain anti-nutrient compounds, potentially affecting their digestibility compared to meat proteins. In this context, this study aimed to compare the nutritional profile and *in vitro* protein digestibility of beef burger (FC), hybrid beef burgers (FH), and plant-based analogue ones (FA and FAF).

II. MATERIALS AND METHODS

Four treatments were elaborated in three batches, as shown in Table 1. The burger samples were prepared by mixing all ingredients in a planetary mixer for 4 minutes. The samples, approximately 100 g each and 12 cm diameter, were immediately frozen at -20°C. The burgers were grilled for 3 minutes on each side at 180°C on an electric grill. Essential amino acids in the burger samples and free amino acids in the digested samples were quantified according to the methodology adapted from previous studies [1]. The in vitro protein digestibility was evaluated according to the Infogest protocol [2] and detailed in a previous study [3]. Differences between treatments were evaluated using one-way ANOVA and the post-hoc Tukey's test with 95% confidence in SPSS software.

Table 1 – Formulations (g/100g) of beef, hybrid, and plant-based analog burgers

Ingredients	Treatments			
	FC	FH	FA	FAF
Lean beef	75	37.5	-	-
Hydrated pea textured protein[1]	-	37.5	75	75
Pork backfat[2]	10	5	-	-
Vegetal fat[3]	-	5	10	10
Methylcellulose	-	-	1.5	0
Flaxseed fiber: psyllium husk (1:1 w/w)	-	-	-	5
Water	13.15	13.15	11.65	8.15

All treatments were elaborated with 1.2% NaCl and 0.6% condiment mix. [1]71.5% moisture, 22% protein, and 1.9% fat. [2] 40% saturated fat. [3]blend of vegetal fat and canola oil (1:1 w/w – 37% saturated fat).

RESULTS AND DISCUSSION

The essential amino acid profiles of the burger samples varied significantly. Analog burgers exhibited the highest levels of calcium, iron, copper, manganese, and magnesium. As expected, the total content of essential amino acids was highest in the beef burger, followed by the hybrid and analogue burgers. The *in vitro* protein digestibility scores also followed this pattern, with the beef burger demonstrating the highest value, followed by FH and FA, and FAF, which showing the lowest protein digestibility, as

shown in Figure 1a. Corroborating these results, the release of free essential amino acids was higher in the beef and hybrid burgers, while the FAF burger showed the lowest value, as illustrated in Figure 1b. Confocal microscopy of the digested samples (Figure 1c) revealed that analogue burgers contained larger protein fragments compared to both beef and hybrid burgers. This suggests that dietary fibres, particularly insoluble ones, may inhibit enzyme activity, thereby reducing protein digestibility. Interestingly, in the FA treatment, a higher number of fat globules was observed (Figure 1c). Methylcellulose, due to its high emulsifying capacity combined with the fact that it is not digested by digestive enzymes probably acted as a barrier to lipid digestibility.

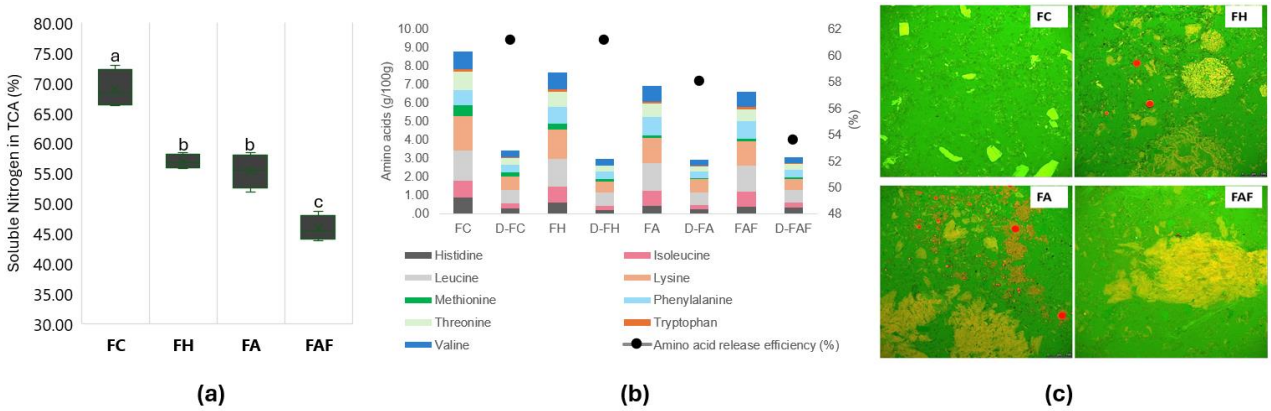

Figure 1. (a) *In vitro* protein digestibility scores based on soluble nitrogen in trichloroacetic acid (10%); (b) Essential amino acids content in the burger samples and in the digested samples (free amino acids) after *in vitro* digestibility protocol and (c) confocal microscopy of digested samples.

III. CONCLUSION

We concluded that pea-based analogue burgers exhibit a good amino acid profile; however, as expected, the level of methionine and the total of essential amino acids was higher in the beef burger. Regarding the *in vitro* protein digestibility, it was higher in the beef burger and lower in the fibre-rich analogue burger. Qualitatively, the analogue burger made with methylcellulose also exhibit reduced lipid digestibility.

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